

Supplementary Figures

Fig.S1

siRNA mediated down regulation of caveolin-1 and caveolin-2 expression in SKOV-3 cells. (A) Western blot analysis of caveolin-1 protein expression in SKOV-3 cells showing, caveolin-1 reduction in siRNA transfected cell lysate (lane1) caveolin-2 expression in transfected cell lysate (lane 2) control siRNA for caveolin-1 (lane 3) and endogenous caveolin-1 expression in SKOV-3 (lane 4). (B) Western blot analysis of caveolin-2 siRNA inhibited protein expression showing, caveolin-2 reduction in siRNA transfected cell lysate (lane1), caveolin-1 expression in transfected cell lysate (lane2), control siRNA for caveolin-2 (lane 3) and endogenous caveolin-2 expression in SKOV-3 (lane 4). β -Actin protein levels were monitored for equal loading between individual lanes. Inhibition of caveolin-1 and caveolin-2 by siRNA resulted reduced EC-eGFP endocytosis in SKOV-3 cells. (C-D) caveolin-1 and 2 and control siRNA transfected SKOV-3 cells were incubated with EC-eGFP at 37°C for 90 min to monitor endocytosis. Images were taken using confocal microscope. Scale bar: 20 μ m.

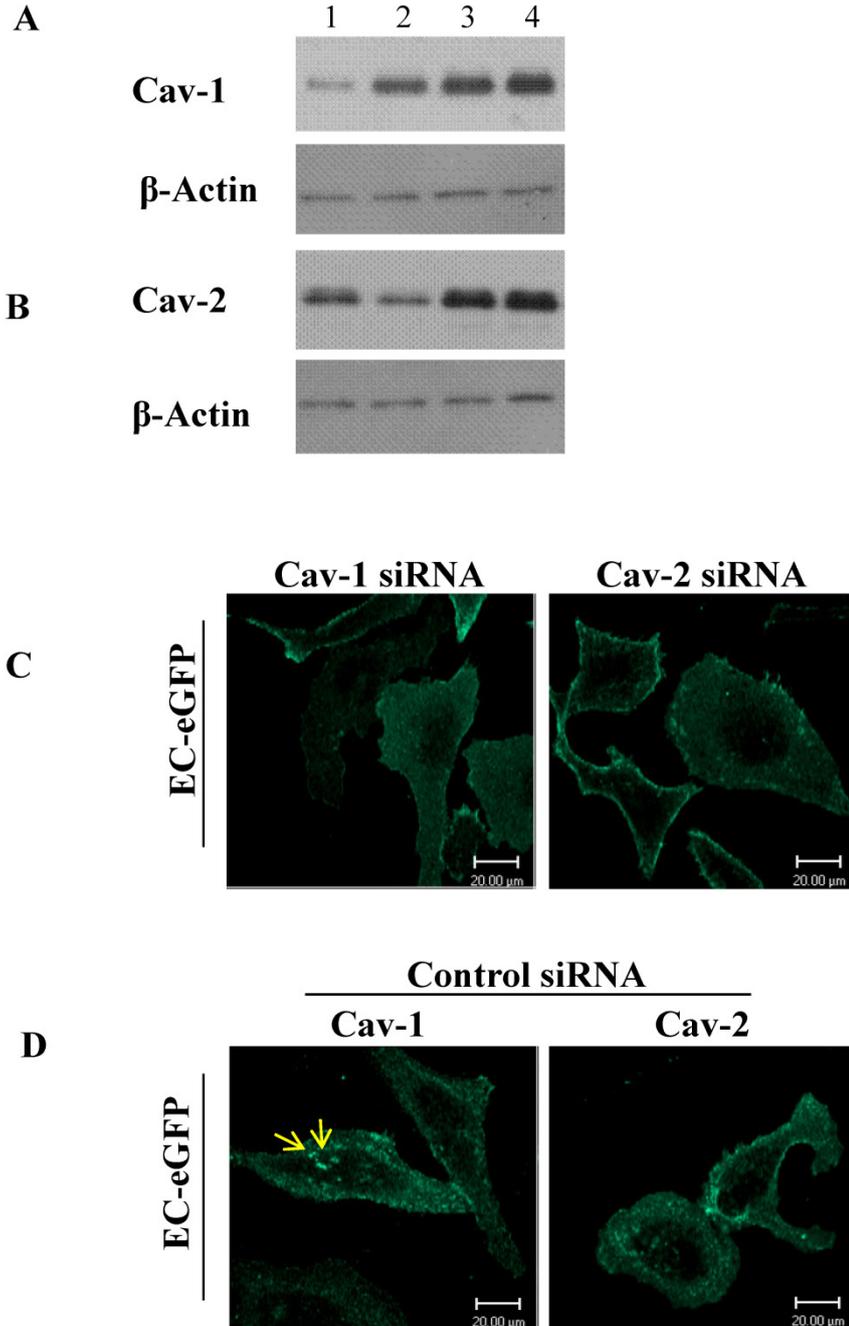


Fig.S2

Caveolin-1 was located intracellularly as small or large puncta in SKBR-3/Cav-1 and SKOV-3 cells. (A) SKBR-3/Cav-1 cells grown in 18 mm cover slips were fixed with 4% PFA, permeabilized in 0.1% Triton X-100, and incubated with anti-caveolin-1 antibody (1:500) for 1 h. Cells were further stained against rabbit IgG labeled with Alexa 555 (1:200) and observed under confocal microscope. Scale bar: 10 μ m. **(B)** Confocal image of caveolin-1 in SKOV-3 cells as explained above showing intracellular puncta.

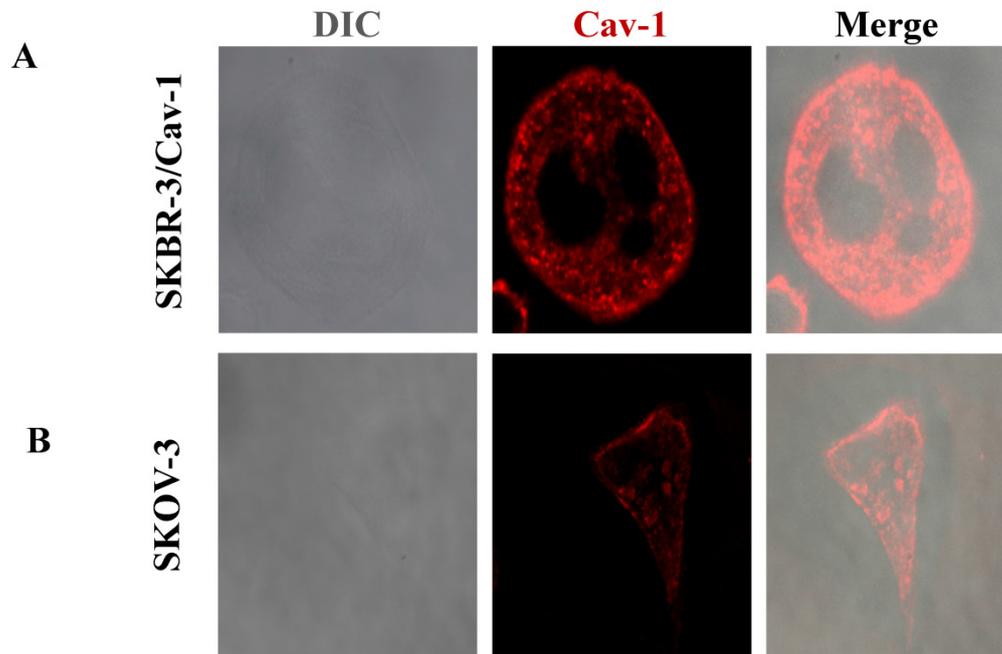


Fig.S3

eGFP was used as a negative control for internalization studies in SKBR-3/Cav-1. Cells were incubated with eGFP at 37°C for 90 min, fixed with 4% PFA, permeabilized using 0.1% Triton X-100, and incubated with anti-caveolin-1 and ErbB2 antibodies (1:500) for 1 h. Cells were further stained against mouse IgG labeled with Alexa 555 (1:200) and rabbit IgG labeled Alexa 350 and observed under confocal microscope. Scale bar: 20 μm.

